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SOLUBLE AND INSOLUBLE DIETARY FIBER IN COOKED COMMON BEAN (*PHASEOLUS VULGARIS*) SEEDS

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Abstract

The common bean (*Phaseolus vulgaris*) requires cooking for extended periods of time prior to consumption. In this investigation both quantitative and microstructural changes in common bean dietary fiber as a result of cooking were examined. Cooking resulted in a slight decrease in soluble dietary fiber and a marked increase in insoluble dietary fiber. The increase in insoluble dietary fiber was responsible for a 15-30 percent increase in total dietary fiber.

Scanning electron microscopy was used to examine the microstructure of uncooked and cooked bean flours and the insoluble and soluble dietary fiber fractions of these two flours. In uncooked whole bean flour large (10-30 μm) spherical starch granules and small (1-5 μm) protein bodies characteristic of the common bean were observed. However, after cooking, only amorphous material containing gelatinized starch and denatured proteins was visible.

Few microstructural differences were observed between uncooked and cooked insoluble dietary fiber fractions. Both fractions consisted primarily of cell wall remnants from which starch and protein storage bodies had been removed. Also present in both insoluble fiber fractions were partially digested fragments of the seed coat palisade cell layer, and long, thin fibers which appear to be remnants of the nutrient transporting phloem. The cooked and uncooked soluble dietary fiber fractions were microstructurally similar consisting of thin, irregularly shaped sheets and long, thin rods.

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Key words: common bean, insoluble dietary fiber, soluble dietary fiber, total dietary fiber, scanning electron microscopy, seed microstructure, autoclaving, cell walls, seed coat, palisade cells, phloem.

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Introduction

The importance of dietary fiber in human nutrition has received a great deal of attention in recent years from scientists and consumers alike (Olson et al., 1987). Numerous health benefits have been associated with consuming adequate amounts of dietary fiber including lower blood cholesterol, reduced risk of heart disease, increased fecal bulk, decreased intestinal transit time, reduced risk of colon cancer, and improved glucose tolerance which is especially beneficial for diabetics (Schneeman, 1986; Toma and Curtis, 1986). Special interest has been focused on common bean dietary fiber because of its high content of metabolically active soluble dietary fiber and its effectiveness in lowering blood cholesterol (Anderson et al., 1984). Fig. 1 shows the insoluble and soluble dietary fiber profiles of the common bean and several commonly consumed cereal products (Dreher, 1987).

Although substantial research has been done on the chemical composition of dietary fiber, considerably less is known about the structural characteristics of dietary fiber. Scanning electron microscopy (SEM) has been used previously to examine the structure of dietary fiber, but this research has focused almost exclusively on insoluble dietary fiber. Insoluble dietary fiber from wheat (Mares and Stone, 1973), wheat bran (Moss and Mugford, 1986; Saunders et al., 1972), rice (Shibuya et al., 1985), corn and soy hull bran (Dintzis et al., 1979), and oat bran (Cadden, 1987) have all been examined with SEM. In contrast, little is known about the structural characteristics of soluble dietary fiber. Several commercially available soluble dietary fiber sources have been examined (Cadden, 1987), but information about the structural characteristics of naturally occurring soluble dietary fiber is lacking. Also, with the exception of some work on soybeans, research on the structural characteristics of dietary fiber has focused primarily on cereals rather than legumes (Wisker et al., 1985). Legume seeds typically contain more dietary fiber than cereals and are a better source of metabolically active soluble dietary fiber.

One area of current research interest is the effect of various forms of processing, including thermal processing, on the dietary fiber content of foods. The effect of various types of cooking on the dietary fiber content of wheat flour (Björck et al., 1984; Siljestrom et al., 1986; Varo et al., 1983), whole wheat flour (Björck et al., 1984; Varo et al., 1983), whole grain wheat (Siljestrom et al., 1986), and potatoes (Dreher et al., 1983; Varo et al., 1983) has been reported. Varo et al. (1983) reported the results of an interlaboratory study where six

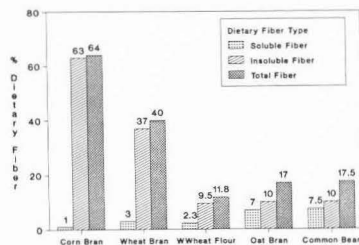


Fig. 1. Comparison of the insoluble and soluble dietary fiber of selected cereal products and the common bean. Values for dietary fiber of cereal products from Dreher (1987).

different laboratories analyzed wheat flour, and whole wheat flour which received one of three treatments: no cooking, normal cooking and cooking under severe conditions. The dietary fiber content of potatoes which were boiled, pressure cooked or French fried was also studied (Varo et al., 1983). Several laboratories participating in the study reported that total dietary fiber in both wheat and whole wheat flour increased as a result of cooking under severe conditions, while other laboratories reported decreases in total dietary fiber using the same samples and cooking conditions. Varo et al. (1983) concluded that measuring changes in the dietary fiber content of foods during cooking is complicated by variability between different analytical methods. Björck et al. (1984) reported significant increases in the total dietary fiber content of extruded whole wheat flour as well as slight increases in the total dietary fiber content of wheat flour when it was extruded under severe conditions. Additionally, a redistribution of insoluble to soluble dietary fiber was observed by Björck et al. (1984) in all extruded wheat flour samples. Siljeström et al. (1986) examined changes in dietary fiber and starch in white wheat flour that had been drum-dried or extruded and whole grain wheat which had been autoclaved, popped or steam flaked. Significant changes in total dietary fiber as a result of cooking were observed only in heat treated whole grain wheat. The total dietary fiber content of whole grain wheat decreased when the grains were either extruded or popped under severe conditions (Siljeström et al., 1986).

Both Varo et al. (1983) and Dreher et al. (1983) examined the effects of different cooking processes on potato dietary fiber. Total dietary fiber increased in potatoes with all forms of cooking investigated including baking, boiling, pressure cooking and the commercial preparation of French fries and potato chips. Soluble dietary fiber content in potatoes was largely unaffected by cooking, with the increase in total dietary fiber resulting from increases in insoluble dietary fiber (Varo et al., 1983; Dreher et al., 1983).

No data are available on the effect of cooking on bean dietary fiber. Cooking is particularly important in the preparation of beans because they contain several heat labile antinutrients (Liener, 1962) and slow swelling starch granules (Thorne et al., 1983; Wursch et al., 1986).

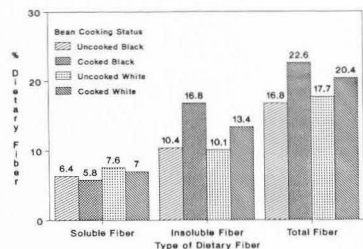


Fig. 2. Changes in the soluble, insoluble and total dietary fiber in white and black common beans as a result of cooking.

Cooking times longer than those needed for cereals are generally required by beans in order to inactivate the heat labile antinutrients and allow for adequate swelling of the starch.

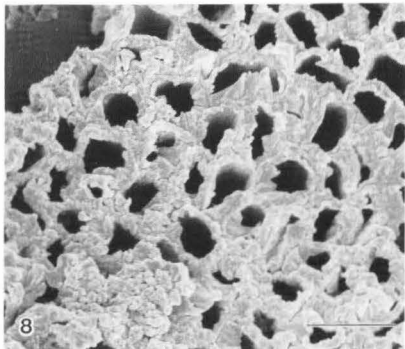
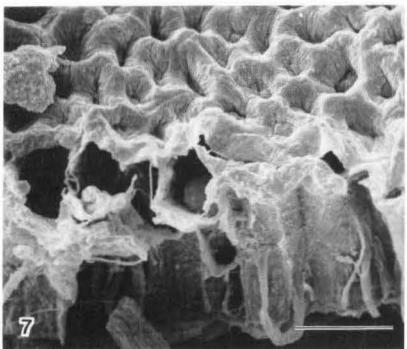
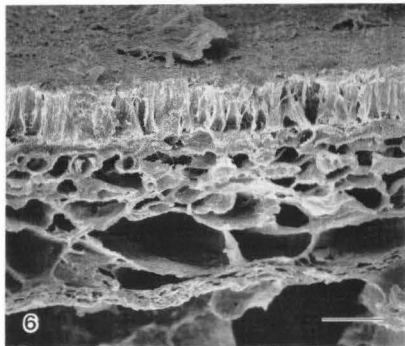
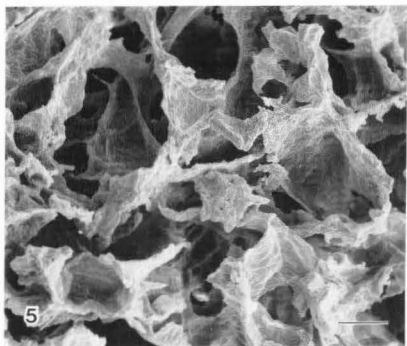
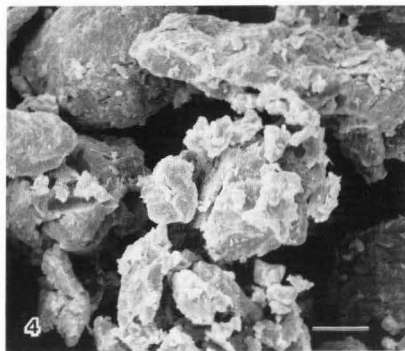
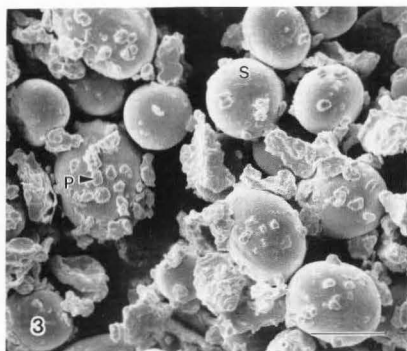
The purpose of this research was to measure changes in common bean dietary fiber content with cooking and to use SEM to observe microstructural changes in the insoluble and soluble dietary fiber fractions as a result of cooking.

Materials and Methods

Common bean (*Phaseolus vulgaris*) seeds with a black seed coat (cv. Tamazulapla) were obtained directly from the producer in the State (Department) of Jutiapa, Guatemala shortly after the 1988 harvest. White beans from the 1988 harvest were purchased shortly after harvest from a seed warehouse in Guatemala City, Guatemala. Both black and white beans were analyzed for changes in dietary fiber content as a result of cooking, however, only black beans were examined with SEM. Beans were cooked in water (1:3) by autoclaving for 20 min at 15 psi and 121°C, and dried overnight in a circulating air oven at 60–70°C. Bean flours were obtained by separately milling uncooked and cooked beans to pass through a 60 mesh screen. The insoluble and soluble dietary fiber content of the resulting flours was determined using the procedure of Asp et al. (1983).

Figs. 3 & 4. Microstructure of whole bean flours. Fig. 3 shows uncooked whole bean flour with both starch granules (S) and protein bodies (P) being present. Fig. 4 shows autoclaved whole bean flours in which the starch granules and protein bodies are no longer distinguishable. Fig. 3, bar = 20 µm; Fig. 4, bar = 50 µm.

Figs. 5–8. Microstructure of uncooked and cooked common bean insoluble dietary fiber. Fig. 5 shows the cell wall remnants typically found in both cooked and uncooked insoluble dietary fiber. Figs. 6, 7 & 8 show the seed coat and palisade cell layers present in the insoluble dietary fiber fraction. Fig. 6 shows an uncooked seed coat which remains largely intact, Fig. 7 shows a cooked palisade cell layer and Fig. 8 shows the interior or lower surface of a cooked palisade cell layer. Figs. 5 and 6, bar = 50 µm; Figs. 7 and 8, bar = 10 µm.



Bean flour was initially gelatinized with termamyl (100° C, 20 min), a heat stable alpha-amylase, and then digested with pepsin (40° C, 60 min) and pancreatin (40° C, 60 min) to remove protein and starch (Prosky et al., 1984). The digestion mixture was filtered to obtain insoluble dietary fiber. Four volumes of 95% ethanol at 60° C was added to the supernatant to precipitate soluble dietary fiber. After one hour soluble dietary fiber was also separated by filtration.

Black bean flour for analysis by SEM was obtained by modification of the procedure of Asp et al. (1983). Insoluble and soluble dietary fiber residues for viewing by SEM were obtained by centrifugation instead of filtration. Insoluble dietary fiber was separated by centrifuging for 30 min at 4000 rpm and freeze dried. Soluble dietary fiber was obtained by centrifuging for 40 minutes at 4000 rpm after ethanol precipitation. Precipitated soluble dietary fiber was initially dried overnight in a vacuum oven at 20° C to remove residual ethanol, and redissolved in distilled water and freeze dried. Dried whole bean flour, and insoluble and soluble dietary fiber samples for examination by SEM were mounted on aluminum stubs and sputter coated with gold (Hummer-Technics). All samples were viewed and photographed at 20 kV with a Hitachi S-570 Scanning Electron Microscope.

Results and Discussion

Cooking and dietary fiber content.

The effect of cooking on the soluble, insoluble and total dietary fiber content of the two bean cultivars examined is shown in Fig. 2. In both black and white beans, cooking resulted in a slight decrease in soluble dietary fiber and a marked increase in insoluble dietary fiber. The increase in insoluble fiber resulted in an increase in total dietary fiber of approximately 15 percent in white beans and 30 percent in black beans (Fig. 2). Other researchers looking at the effect of thermal processing on changes in dietary fiber content of wheat and potatoes have reported either no change (Varo et al., 1983) or increases in insoluble fiber with no change in soluble dietary fiber (Björck et al., 1984; Dreher et al., 1983; Varo et al., 1983). Changes in common bean dietary fiber with cooking most closely resemble changes observed previously in potatoes where insoluble dietary fiber increased and soluble fiber remained largely unchanged regardless of the type of cooking (Dreher et al., 1983; Varo et al., 1983).

The chemical basis for changes in the dietary fiber content of foods during cooking remain unclear. The formation of resistant starch (Björck et al., 1986), amylose-lipid complexes and Maillard-reaction products (Björck et al., 1984) have been hypothesized as contributing to observed increases in dietary fiber. However, amylose-lipid complexes appear to be digestible *in vivo* (Holm et al., 1983), and in a low lipid food like the common bean, formation of amylose-lipid complexes is not likely to contribute significantly to changes in dietary fiber as a result of cooking. Additional research on the formation of resistant starch and Maillard-reaction products during cooking is needed to determine their contribution to changes in dietary fiber content.

Microstructure of whole bean flour.

Structural differences between uncooked and cooked common bean flours are shown in Figs. 3 and 4. Present in the uncooked flour are large (10-30 μ m),

spherical starch granules and smaller (1-5 μ m) protein bodies similar to those previously observed in intact seeds (Hughes and Swanson, 1985; Swanson et al., 1985). In the cooked flour, starch granules have been gelatinized and protein bodies denatured leaving primarily amorphous material that is irregular in size and shape (Fig. 4).

Microstructure of insoluble dietary fiber.

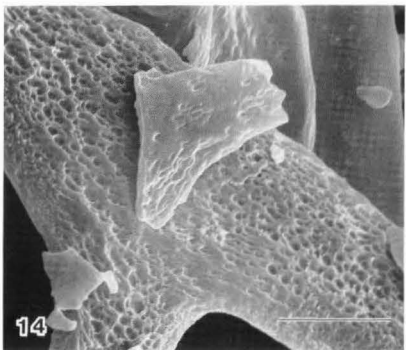
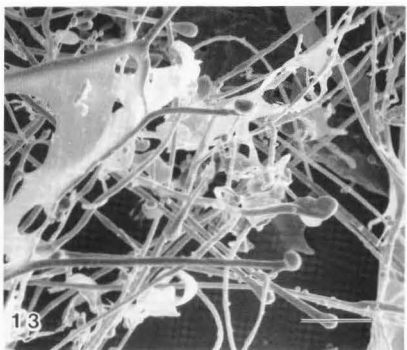
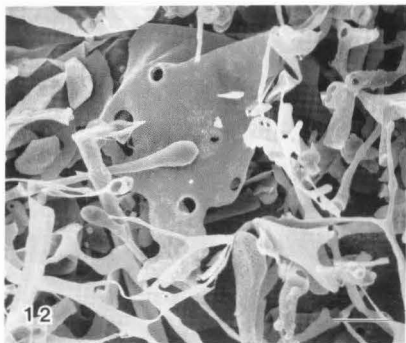
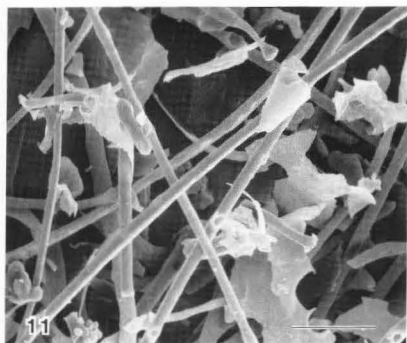
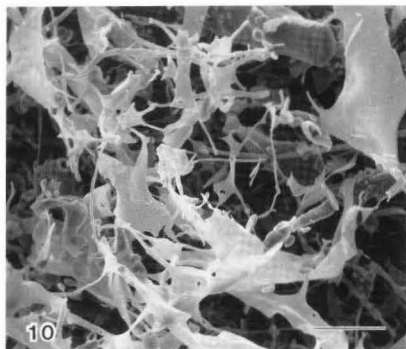
The microstructure of cooked and uncooked common bean insoluble dietary fiber is shown in Figs. 5-9. Common bean insoluble dietary fiber is composed primarily of cellulose and hemicelluloses (Selvendran, 1984). Few differences were observed between the uncooked and cooked insoluble dietary fiber fractions. Structural differences may have been minimized by gelatinization of the uncooked sample for 20 min at 100° C during the first stage of digestion (Asp et al., 1983). Both uncooked and cooked insoluble fiber consisted primarily of cell wall remnants from which all starch and protein storage bodies had been removed (Fig. 5). Also visible were undigested portions of the seed coat, with the long cylindrical cells of the seed coat palisade cell layer (Fig. 6). The exterior surface of the palisade cell layer in uncooked fiber was relatively flat (Fig. 6) and similar to exterior surfaces previously observed in unimbibed whole seeds (Swanson et al., 1985). However, examination of the outer surface of the palisade cell layer of cooked insoluble dietary fiber revealed a rolling, uneven surface (Fig. 7). The uneven surface appears to have been caused by differential swelling of the palisade cells during cooking.

When viewed from the exterior surfaced or in cross-section, palisade cells in both uncooked and cooked insoluble dietary fiber appear intact. However, examination of the interior or lower surface reveals that the lower surface of the palisade cells was removed, presumably by enzymatic digestion during preparation of the dietary fiber (Fig. 8). Removal of the lower surface of the palisade layer allowed the internal contents of the cells to be digested and removed. Thus, while palisade cells in insoluble dietary fiber appear unaffected by digestive enzymes, these cells are actually empty chambers or lumens from which all internal contents have been removed. Similar palisade cell chambers were observed by Dintzis et al. (1979) when examining digested soy bean hulls.

Long, thin fibers (Fig. 9) were an unusual feature of insoluble dietary fiber not observed in other fractions. These fibers were approximately 10-15 μ m wide and coarse in appearance (Fig. 9). Though the exact origin of the fibers is uncertain, they appear morphologically

Fig. 9. Long, thin fibrous material present in the uncooked insoluble dietary fiber believed to be part of the phloem system. Bar = 25 μ m.

Figs. 10-14. Microstructure of uncooked and cooked common bean soluble dietary fiber. Figs. 10 and 11 show the sheets and rods typically observed in uncooked soluble dietary fiber while Figs. 12 and 13 show the same features for cooked soluble dietary fiber. Fig. 14 shows a rod-like structure present in the soluble dietary fiber at greater magnification so that the porous nature of the material is evident. Figs. 10 and 13, bar = 50 μ m; Figs. 11 and 12, bar = 25 μ m; Fig. 14, bar = 10 μ m.



similar to the nutrient transporting phloem previously observed in whole seeds (Hughes and Swanson, 1985).

Microstructure of soluble dietary fiber.

The microstructure of the cooked and uncooked soluble dietary fiber fractions is shown in Figs. 10-14. Common bean soluble dietary fiber is composed primarily of pectic substances (Selvendran, 1984). Microstructurally, common bean soluble dietary fiber appears as thin, irregularly shaped sheets (Figs. 10 and 11) attached to a framework of long thin rods (Figs. 11, 12 and 13) in both uncooked and cooked samples. Closer examination of the rods observed in the soluble fiber (Fig. 14) revealed that they were thinner (2-4 μm) and less fibrous in nature than the fibers observed in the insoluble fiber (Fig. 9). Many of the rods also exhibited pods or bulges at one end (Figs. 12 and 13). The soluble dietary fiber structures observed in this investigation are very different from the structures observed by Cadden (1987) in samples of commercially available soluble dietary fiber. The structures reported by Cadden (1987) are similar to structures observed previously (Hughes and Swanson, unpublished data) of soluble dietary fiber after ethanol precipitation and before being redissolved in water and freeze dried. Differences between our observations and those reported by Cadden (1987) probably result from different preparation procedures and should not be interpreted as significant structural differences in soluble dietary fibers from different sources. SEM examination provides useful information on the structural characteristics of soluble dietary fiber. However, it is important to remember that in its natural state the fiber is solubilized in water and structural artifacts may result from the extensive dehydration required.

Conclusions

The cooking of common bean flours resulted in marked increases in insoluble dietary fiber and total dietary fiber while soluble dietary fiber content decreased slightly. In uncooked whole bean flour, SEM revealed starch granules and protein bodies characteristic of the common bean while cooked flour contained amorphous material consisting of gelatinized starch and denatured proteins. Microstructural examination revealed common bean insoluble dietary fiber consisting primarily of cell wall remnants and portions of the seed coat palisade cell layer. Long thin fibers believed to be remnants of the nutrient transporting phloem were also observed. The common bean soluble dietary fiber, in contrast, consists of thin, irregularly shaped sheets and long, thin rods. No significant microstructural changes in either the insoluble or soluble dietary fiber fractions were attributed to cooking. Structural differences between uncooked and cooked may have been minimized by the need to digest uncooked samples for 20 min at 100°C.

Changes in the dietary fiber content of foods with cooking is a complex and poorly understood phenomenon influenced by the analytical method used, the type of food studied, and the type, duration and severity of cooking utilized. Additional research on the chemical processes responsible for quantitative changes in dietary fiber as a result of cooking will clarify the relative contribution of each of these variables.

Acknowledgements

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Discussion with Reviewers

W.J. Wolf: Since the soluble fiber fraction was freeze-dried, one must be concerned about possible artifacts, particularly the sheet-like materials. If the sheet-like materials pre-existed, is it likely that the fiber would have been soluble?

Authors: As indicated in the paper, the examination of a water soluble material in its dehydrated state has numerous drawbacks. It would seem reasonable to assume that rods would be more water soluble than sheet-like material, and that the sheets were formed during dehydration as a result of the agglomeration of rods. However, we have no evidence to support or contradict such a hypothesis.

F.R. Dintzis: Were both soluble and insoluble dietary fiber fractions starch and/or protein free?

Authors: The residual protein content of all dietary fiber fractions was determined according to the method of Asp et al. (1983) and protein was found in all fractions in widely varying quantities (3-25% of total protein). We did not examine the bean dietary fiber fractions for starch, but resistant starch has been reported in cereal dietary fiber (Björck et al., 1986) and could logically be assumed to also be present in common bean dietary fiber.

G.L. Hosfield: How many seeds were examined before a particular photomicrograph was chosen to represent the corresponding SEM observations as a figure?

Authors: Three to five samples of each of the flours or dietary fiber fractions were examined prior to selecting a representative micrograph. Our experience has been that thorough examination of a few samples is more fruitful in obtaining representative micrographs than cursory examination of a large number of samples.

G.L. Hosfield: From a human nutritional viewpoint, what is the significance of the dietary fiber research results and conclusions?

Authors: Two nutritional implications seem readily apparent. First, in foods such as beans which are rarely if ever consumed raw, data on the dietary fiber of cooked beans is more valuable than data on raw beans. Second, though both insoluble and soluble dietary fiber are desirable in the diet, their metabolic effects are different. Therefore, any shifts in soluble and insoluble dietary fiber as a result of cooking should be included in food databases in order to more accurately represent the expected metabolic effects of the dietary fiber present in the food.

G.L. Hosfield: What role does the bean seed coat play in fiber quantity and quality?

Authors: The seed coat of the common bean typically comprises 8% of the whole seed by weight. The seed coat is also typically high in insoluble dietary fiber and low in soluble dietary fiber.

A.C. Olson: The SEM was only done on the black bean which is not a major bean of commerce in the United States. How widely is this bean an item of commerce?

Authors: Though black beans are not widely cultivated or consumed in the United States, they are widely produced and consumed throughout Latin America. In several Latin American countries including Guatemala, black beans are the preferred bean and are consumed on a daily basis. The particular cultivar (Tamazulapa) investigated in this study, however, is not to our knowledge grown and consumed outside of Guatemala.

A.C. Olson: What effect (if any) do you think the heat treatment during digestion with termamyl and the milling had on your results?

Authors: The need to digest the uncooked flour for 20 min at 100°C probably minimized microstructural differences between cooked and uncooked dietary fiber fractions and may have also reduced quantitative differences. Foods are routinely milled prior to determination of dietary fiber content. However very finely milled samples of a food have been shown to have a lower insoluble dietary fiber content than coarsely milled samples. For this reason it is important to report the size to which the food was milled prior to dietary fiber determination.